Proc. Indian Acad. Sci., Vol. 87 B, No. 9, (Animal Sciences-3), September 1978, pp. 261–266,

Growth and protein efficiency ratio in albino rats—a statistical approach

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MS received 7 July 1978

Abstract. In animal experiments the Randomised Block Design is quite often used, the blocking based on some characteristics related to the character under study. Evaluation of the relative efficiency of the Randomised Block Design as compared to completely Randomized Design is necessary to decide whether a change of design is warranted. Data from 465 growth and/or protein efficiency ratio (PER) studies were processed to evaluate the relative efficiency of the randomised block design. The randomised block design turned out to be more efficient in growth and PER studies when blocking is based on age, sex and initial weight.

Keywords. Growth; protein efficiency ratio; completely randomized design; randomized block design; relative efficiency.

1. Introduction

In designing an experiment on statistically sound principles, there is every opportunity for ingenuity in the method of attacking the problem so that it fulfils the objectives of the experiment. An experiment having a specific objective may be designed in two or more quite different ways; with the same cost in terms of experimental effort, one may lead to unambiguous results, whereas the others may lead to ambiguous results, no matter what the outcome is. How good a design is in comparison with others for handling the same basic problems may be measured in terms of (i) the relative cost of the experimental effort, and (ii) the relative precision with which conclusions may be stated.

Contrary to the general belief more precise conclusions do not always demand greater experimental effort, but more careful attention to experimental design. Even the most sophisticated analysis of data from a poorly designed experiment may not fulfil its requirements whereas an imperfect or inefficient analysis of the data from a well designed experiment may give conclusions that are enlightening and substantially correct.

Over the past several years at Central Food Technological Research Institute, the nutritive values of numerous food formulations are being assessed through animal feeding experiments using the Wistar strain albino rat as the experimental animal. Invariably, the Randomized Block Design (RBD) has been adopted using sex, age and initial weights of the rats as the criteria for forming blocks, as the gain in weight is said to be influenced by these factors.

The animals that are being used for experimentation are inbred and hence could be considered as uniform for all practical purposes. RBD under these circumstances, theoretically, would mean little more than a random procedure and CRD can be used without any disadvantage.

In this paper an attempt has been made to evaluate statistically the efficiency of the RBD over CRD and decide whether it is worthwhile to continue using the RBD or a switch over to the CRD is warranted.

2. Materials and methods

Initially, the study has been restricted to growth and protein efficiency ratio (PER) experiments. Data from 465 growth and PER experiments carried out on (male female) wistar strain albino rats have been processed.

From the analysis of variance (ANOVA) of each experiment, E_e (CR), the error variance, if CRD had been used instead of RBD was calculated by using the expression:

$$E_e\,(CR) = \frac{n_b\,E_b + (n_t + n_e)\,E_e}{n_b + n_t + n_e}$$

where E_b, E_e are the block and error mean squares, n_b, n_t and n_e are the degrees of freedom for block, treatment and error respectively from the ANOVA of RBD (Steel and Torrie 1960).

The efficiency of RBD relative to CRD was computed as a percentage from the expression

$$RE\% = \frac{E_e\,(CR)\,(n_1 + 1)\,(n_2 + 3)}{E_e\,(RB)\,(n_2 + 1)\,(n_1 + 3)} \times 100$$

where E_e (CR) and E_e (RB) refer to the error mean squares under CRD and RBD respectively and $n_1 = n_b$ and $n_2 = n_b + n_e$ (Steel and Torrie 1960).

Comparing the amount of information available from a RBD with t treatments and b blocks, i.e., $t \times b$ units, and the number of units necessary to get the same amount of information using a CRD given by $t \times b \times RE\%/100$, the savings or loss in terms of experimental units, viz., rats, was calculated for each experiment.

Wilcoxon's test (Steel and Torrie 1960) was used to determine the significance of the difference between average savings and average losses.

3. Results and discussion

The frequency distribution of the number of experiments grouped on the basis of their $RE\%$, sex-wise for growth and PER studies as well as their break-up according to whether their $RE\%$ is greater than 100, or less than or equal to 100 has been presented in table 1a and 1b. It is evident from these tables that the relative efficiency of the RBD is >100 in more than 57% of the experiments in growth studies of both sexes. As regards PER, 50% of the experiments with males and 55% with females fall in this group.

Table 1a. Frequency distribution of the experiments according to their RE%.

RE%	Growth		PER	
	Males	Females	Males	Females
70- 80	8	8	4	4
80- 90	17	12	27	9
90-100	41	44	56	18
100-110	30	31	45	15
110-120	19	14	21	14
120-130	14	6	13	3
130-140	7	12	3	6
140-150	5	8	1	—
>150	14	16	5	—
Total	155	151	175	69
Means	108·8	110·4	102·9	103·6

Table 1b. Classification according to RE < 100 or >100 with the percentages

RE%	Growth		PER	
	Males	Females	Males	Females
<100	66(42·6)	64(42·4)	87(49·7)	31(44·9)
>100	89(57·4)	87(57·6)	88(50·3)	38(55·1)

Further the mean *RE%* in the case of growth studies is 109 and 110 for males and females respectively whereas for PER studies it is about 103 irrespective of sex.

Table 2 presents the percentage of experiments with *RE%*>100 and *RE%*≤100 classified according to the number of treatments and the average number of experimental units saved or lost for getting the same amount of information by using RBD instead of CRD.

In the case of growth studies with males the RE% is >100 in more than 50% of the experiments with differing number of treatments except when the number of treatments is 5. Even in the case when the number of treatments is 5 and when only 46·7% of the experiments have efficiency >100, the average savings per experiment in terms of experimental units is 11 while the number of losses from the rest 55.3% of the experiments with *RE%*<100 is only 3.

The growth studies in females reflect the fact that more than 50% of the experiments with treatments more than 2 have *RE%*>100; while with only two treatments 33·3% of the experiments have *RE%* >100. The average savings from the 33·3% being 15 while the average losses from the rest 66.7% of the experiments being 2.

As regards PER studies with males when the number of treatments are 4, 6, 8 and ≥9 more than 50% of the experiments have *RE%*>100. In the other cases of 2, 3, 5 and 7 treatments the percent of experiments having *RE%*>100 is less than 50. But the average savings in all the cases exceed the average losses.

In the case of females except where the number of treatments is three in all other cases more than 50% of the experiments have *RE%*>100. The average savings being 6 and the average losses 3.

Table 2. Per cent of experiments with relative efficiency > 100 and relative efficiency ⩽ 100, number of treatment-wise along with the respective average savings and losses in terms of experimental units.

No. of treatments	Growth								PER							
	Males				Females				Males				Females			
	% with RE >100	Av. savings per expt.	% with RE ⩽100	Av. losses per expt.	% with RE >100	Av. savings per expt.	% with RE ⩽100	Av. losses per expt.	% with RE >100	Av. savings per expt.	% with RE ⩽100	Av. losses per expt.	% with RE >100	Av. savings per expt.	% with RE ⩽100	Av. losses per expt.
2	72·2	7	27·8	2	33·3	15	66·7	2	45·4	7	54·6	5	—	—	—	—
3	52·0	6	48·0	3	61·1	8	38·9	3	36·4	3	63·6	3	44·0	4	56·0	3
4	59·3	6	40·7	3	52·4	7	47·6	2	50·0	6	50·0	3	54·5	5	45·5	4
5	46·7	11	53·3	3	80·0	7	20·0	4	25·0	9	75·0	3	66·7	3	33·3	4
6	52·2	4	47·8	3	57·9	8	42·1	2	73·1	5	26·9	4	80·0	7	20·0	3
7	75·0	8	25·0	2	62·5	12	37·5	3	37·5	8	62·5	3	70·0	9	30·0	3
8	83·3	11	16·7	0	50·0	8	50·0	1	78·9	4	21·0	4	60·0	11	40·0	3
⩾9	54·5	10	45·5	3	59·0	4	41·0	3	54·5	5	45·5	3	55·5	6	44·5	3
Mean		8		2		9		3		6		4		6		3

Table 3. Cost analysis

		Mean RE%	Av. No. of units saved/ experiment	Approx. cost/unit Rs.	Savings/ expt. Rs.
Growth	Males	108·8	9	30	270
	Females	110·4	10	30	300
PER	Males	102·9	3	25	75
	Females	103·6	4	25	100

The difference between average savings and average losses has been tested by Wilcoxon's test (Steel and Torrie 1960). The savings are significantly greater compared to losses in all the cases; $P<0{\cdot}01$ for growth studies of both sexes as well as PER of males, $P<0{\cdot}05$ for PER studies of females.

Based on the average savings calculated from the mean *RE*% from table 1a, the cost analysis is presented in table 3. By the use of RBD 9-10 units per experiment are saved in growth experiments and 3–4 in PER experiments irrespective of sex. The approximate cost/unit ranges from Rs 25–30. Annually 150 rat feeding experiments are being carried out at the Institute out of which on an average 100 are growth and/or PER studies. Hence it is presumed that on an average an amount of Rs 18,000 per annum may be saved by continuing to use the RBD.

It is evident from the above study that RBD is more efficient compared to CRD in growth and PER studies. However, it is worthwhile to study the feasibility of increasing its relative efficiency by including additional factors like position and environment while forming blocks, as these factors are known to have an effect on biological response. It was observed in an incidental study (Sankaran, personal communication), that the rats placed in two different rooms differed significantly ($P<0{\cdot}001$) in their growth rate which confirms the presence of an environmental effect. Further in another study on growth (Krishnakumari *et al* 1976), the effect of different positions on the same rack with a number of shelves was observed. The rats placed on the top-most shelf gained more weight as compared to those on the bottom-most shelf.

4. Conclusions

Hence, it can be concluded that with the same amount of experimental effort it would be possible to further increase the relative efficiency of the RBD if factors like position and environment are also taken into account along with other characteristics in forming blocks. This will simultaneously result in greater savings in terms of experimental units and cost.

Acknowledgements

The authors are indebted to Sri A N Sankaran for his valuable comments which aided in improving the quality of the paper. They are also thankful for the encouragement received from Dr V H Potty, Industrial Development and Consultancy Services and Dr B L Amla, the Director of the Institute.

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